

ICP ANALYSIS OF HEAVY METALS IN *MERETRIX* SPP. FROM PANTAI SEPAT, KUANTAN, PAHANG

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ABSTRACT

Hard clams constitute the genus *Meretrix* is one of the most demanded bivalvia species in Malaysia. However, the unknown level of heavy metal content raises concerns for the health of seafood lovers, particularly those who consume seafood from Pantai Sepat, Kuantan, Pahang. Using bivalves as bio-indicators, the objectives of this study are to determine the level of concentration of Copper (Cu), Zinc (Zn), Cadmium (Cd) and Lead (Pb), and to evaluate these concentrations according to the permissible limits established by the Malaysia Food Act (1983) and Food Regulations (1985) and the World Health Organisation (1982). Before analysing the samples with Inductively Coupled Plasma Optical Emission Spectroscopy (ICP-OES), the collected and desiccated samples were digested with acid. The concentrations of copper (Cu), zinc (Zn), cadmium (Cd), and lead (Pb) were determined to be 0.56 ppm, 6.76 ppm, 0.06 ppm, and 0.04 ppm, respectively. The order of metals in the collected *Meretrix* spp. is Zn > Cu > Cd > Pb. The concentrations of all elements present in the hard clams were assessed and found to fall within acceptable thresholds, indicating that they are suitable for consumption without posing any health risks.

Keywords: heavy metal; *Meretrix* genus; Pantai Sepat, Pahang

Introduction

Molluscs, particularly those belonging to the bivalve class, are commonly employed as bioindicators in the field of environmental monitoring. The bivalve organism possesses the necessary characteristics to serve as a potential bioindicator, as it has the capacity to accumulate contaminants from its surrounding environment. Typically, the concentration of pollutants present in the tissue of an organism is employed as a means of evaluating the extent of pollution within its environment (AbdAllah and Moustafa, 2002). According to Ambarwati et al. (2020), there are 15 species that were recorded worldwide known as *M.*

astricta, *M. attenuate*, *M. aurora*, *M. casta*, *M. lamarckii*, *M. lusoria*, *M. lyrata*, *M. meretrix*, *M. morphine*, *M. petechialis*, *M. planisulcata*, *M. subtrigona*, *M. vestita*, *M. tigris*, and *M. marisarabicum*. In addition, 38 species and subspecies comprise the genus *Meretrix*. According to the systematisation, *Meretrix lurosia* is considered a synonym of *Meretrix meretrix* (Xie et al. 2012). In addition, calcium-rich molluscs shells have been used in the poultry industry. It has been demonstrated that calcium oxide produced from *M. meretrix* shells is an effective catalyst for biodiesel production. Not only do these shells serve an important ecological purpose, but they also have commercial value (Ambarwati et al. 2020). The shell of *M. meretrix* has artistic and decorative applications (Xie et al. 2012). Since the majority of species in this genus are edible, such as *M. meretrix* and *M. lyrata*, they are frequently caught and traded.

The Veneridae family of edible bivalves contains a diverse variety of species. *Meretrix* is a genus within the family Veneridae (Ambarwati et al. 2020). *Meretrix* spp. is common edible saltwater mussels that are widely distributed in eastern Asia and contain high levels of nutrition and valuable medicinal properties. It is also a valuable source of Traditional Chinese Medicine (TCM) and a marine sustenance source, and it is commonly known as the Asiatic hard clam. *Meretrix* is ordinarily discovered in estuaries and mangrove habitats (Ambarwati et al. 2020). In addition, the clam typically burrows in nutrient-rich sand and silt sediments in mudflat areas of estuaries (Admodisastro 2021). This mollusc is capable of accumulating heavy metals due to its lifestyle and habits, which serve as bioindicators.

Heavy metals are extremely tenacious, resistant to bacterial contamination, do not deplete, and can cause adverse effects by inducing diverse reactions in animals and vegetation (Ong & Ibrahim 2017). Trace metals and metalloids can infiltrate aquatic ecosystems through natural weathering, geochemical weathering, and human domestic and industrial activities, among others. Plus, marine organisms can accumulate heavy metals such as copper (Cu), iron (Fe), cadmium (Cd), lead (Pb), chromium (Cr), zinc (Zn), and nickel (Ni) (Viera et al. 2021). Since they cannot be degraded biologically, their accumulation in the delicate tissues of bivalves can be harmful to humans who consume these organisms as food sources. Distinct aquatic organisms that are known as filter feeders suspend particles in water that is polluted by a variety of pollutants, such as heavy metals resulting from either human activities or natural emissions. In addition, they have extremely sluggish enzyme systems capable of degrading persistent organic contaminants (Dabwan & Taufiq 2016).

In addition, there are numerous studies on the deleterious effects of heavy metals on aquatic ecosystems and the human health of marine consumers. According to Ambarwati et al. (2020), the concentrations of Pb, Sn, Hg, Cd, and Sb in bivalve samples were found to be lower than the permissible limits recommended by the Malaysia Food Act (1983) and Food Regulations (1985). Consequently, the results indicated that all samples of bivalves are healthy for human consumption. The majority of living organisms require fewer amounts of essential metals such as Fe, Mn, Cu, and Zn for essential processes such as growth; however, if the amounts exceed the standard limit, they will cause hazardous effects. Another study from Mohd Harun, Jovita and Ahmad Zharin (2007) also found that the *M. meretrix* R. collected from North Borneo contained the highest concentration of Cd and Cr among the studied molluscs. In a study conducted by Hung et al. (2001), an examination was made towards the trace metal concentrations in various species of molluscs, including *Meretrix lusoria*, collected from the coastal region of Taiwan. In contrast, Wang et al. (2005) conducted a study on the mollusc species *Meretrix meretrix* to investigate the presence of heavy metal contamination in coastal areas adjacent to the Chinese Bohai Sea. Nevertheless, none of the aforementioned research has particularly examined the biological cumulative factor of such species. The investigation and reporting

of heavy metal concentration levels in *Meretrix meretrix* Roding (*M. meretrix* R.) residing in estuaries have not been previously conducted. *M. meretrix* R. is a bivalve mollusc that inhabits the unconsolidated substrate of aquatic environments and is often distributed in the coastal regions of Borneo Island. These organisms possess the characteristic of being filter feeders, which renders them highly susceptible to the accumulation of hazardous chemicals present in both water and sediment.

Due to high industrial development in Kuantan, the risk of trace metal contamination is currently significant to discover. Therefore, the objectives of this study were to determine the level of concentration of selected heavy metals at Pantai Sepat, Kuantan and to evaluate these concentrations according to the permissible limits established by the Malaysia Food Act (1983) and Food Regulations (1985) and the World Health Organisation (1982).

Materials and Methods

Study area

Latitudes and longitudes (3°42'16.0" N, 103°20'12.0" E) define the sampling area along the coastline of Pantai Sepat, Kuantan Pahang. Pantai Sepat is an ordinary Malay fishing village as well as one of Pahang's most attractive beaches. It takes approximately 20 minutes to reach the royal settlement of Pekan from the centre of Kuantan. *Meretrix* spp. samples were collected on Pantai Sepat in August 2022. The Identification was conducted by observing the exterior and interior of the shells by referring to Dharma (2005) and Huber (2010).

Sample collection

Randomly, thirty *Meretrix* spp. were collected along the coastline. The samples were collected with both a long metal implement and by hand. The long metal scraper was dug into the soil and dragged until it produced a clicking sound when it struck a boulder, indicating the presence of clams. The encrusted organisms and sediment were then removed from the clams by soaking them in seawater for a full night. Then, they were stored at a low temperature in a freezer for one night. The collected clams were then transported to a laboratory for analysis using a cool crate to maintain a low temperature.

Samples digestion

The heavy metal components in the sample were analysed with Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES), the acid digestion method was used in conjunction (Aradhi, Kurakalva and Nyasavajhula 2009). Before beginning the experiment, the glassware and other equipment were cleaned with detergent and then rinsed with distilled water. The beakers were entirely and sterilely dried in the oven. The collected clams were then washed with tap water to remove superfluous sediment from the outer shell. Then, using a ceramic knife, the delicate tissues were dissected from the shell. The samples were transferred into a labelled petri dish and desiccated in an oven at 100 degrees Celsius for 24 hours to achieve a constant weight. The following day, the dried samples were grounded into powder using a porcelain mortar and pestle. The substance was then sieved into a finer powder to facilitate digestion.

Heavy Metal Determination

For the analysis of the heavy metal content in the bivalve samples, the acid digestion method was employed. This technique is used to convert solid samples into liquid samples because the ICP-OES machine can only operate with liquid solutions. The procedure was conducted in the fume hood for safety reasons. A total of 2 grams of dried powdered tissue samples were transferred to a beaker for acid digestion. Then, 2 mL of sulfuric acid and 2 mL of nitric acid were added to the beaker and combined. The samples were then heated for 30 minutes at 60°C on a hot plate. After observing the reaction for thirty minutes, the beaker was covered with a watch glass and cooled. The temperature continued to rise until it reached 150°C after 4 mL of nitric acid was added to the beaker. After the sample had been thoroughly digested, it was allowed to settle at room temperature. Then, 1 mL of 30% hydrogen peroxide was added to the vial for every 30 minutes until a clear solution was achieved. The sample solution was poured through filter paper into a 100-mL Erlenmeyer vial to separate it from the sediment. The solution was subsequently transferred to a 20-mL falcon tube and diluted with deionized water. The sample was refrigerated at 20 degrees Celsius prior to further analysis. The sample was then transferred to a tube and analysed for heavy metal content using ICP-OES.

Results and Discussion

Thirty individuals of the genus *Meretrix* were collected at the sampling site and decomposed using the acid digestion technique. The liquid sample was then analysed using ICP-OES to determine the concentration of four distinct categories of heavy metals, including Cu, Zn, Cd, and Pb.

Table 1 summarises the concentrations of heavy metals in the *Meretrix* genus from Pantai Sepat, Kuantan. According to Table 1, Zn has the highest concentration of the four heavy metals, followed by Cu, Cd, and finally Pb. In addition, the measured concentrations of heavy metals are compared to the permissible limits of heavy metal concentrations in food established by the Malaysia Food Act (1983) and Food Regulations (1985) and the World Health Organisation (1982), as shown in Table 2. The purpose of this comparison is to determine whether the concentration of heavy metals in the *Meretrix* genus falls within or exceeds the permitted limits.

Table 1: The measured concentration of heavy metals in *Meretrix* genus by ICP-OES

Elements	The measured concentration of heavy metals (ppm)
Copper (Cu)	0.56
Zinc (Zn)	6.76
Cadmium (Cd)	0.06
Lead (Pb)	0.04

Table 2: Comparison between the measured concentration of heavy metals in the *Meretrix* genus and the permissible limit of the concentration of heavy metals in food

Elements	The measured concentration of heavy metals (ppm)	The permissible limit of concentration of heavy metals in Food (ppm)
Copper (Cu)	0.56	30.00
Zinc (Zn)	6.76	100.00
Cadmium (Cd)	0.06	1.00
Lead (Pb)	0.04	2.00

Among the four elements listed in Table 1, Zn has the greatest concentration at 6.76 parts per million (ppm). Zinc has accumulated in the hard clams due to its use as an anticorrosion and antifouling agent in marine paint (Chuan & Ibrahim 2017). Given that Pantai Sepat is surrounded by fishing communities, such as boating, transportation, and fishing, it is likely that these trawlers will be worn frequently. According to Yusoff et al. (2021), the marsh clam (*Polymesoda expansa*) has the highest concentration of Zn among Cu, Cd, and Pb. At the sampling site, the Kelantan River, there are numerous mining, transport, and fishing boat activities, according to the study. In addition, there are restaurants that serve seafood to their customers and may dispose of their refuse in the nearby marine system. All of these potential sources may contain zinc, which can leach into aquatic habitats and accumulate in the water column and detritus. The hard clams filter the water to capture food particulates, which could facilitate Zn absorption (Yun-Ru et al. 2020). Nevertheless, according to the article by Chuan and Ibrahim (2017), Zn is an essential element that is required for metabolic activities of living organisms related to nutrition and biochemical pathways, indicating that Zn is essential for living organisms' metabolic processes. Consequently, this may also explain why its concentration is greater than that of non-essential elements. Despite its importance to organisms, it can have toxic effects when consumed in excess or in low concentration over a prolonged period of time (Jaishankar et al 2014).

However, the concentration of Zn in the firm clam is below the permissible limit established by the Malaysia Food Act (1983) and Food Regulations (1985) and the World Health Organisation (1982), as shown in Table 2. The daily permissible limit is 100.00 ppm. Due to the large value difference, it can be concluded that Zn contamination in the *Meretrix* genus is low and the clams are safe to consume. However, the consumption of clams should be monitored periodically, as low concentrations ingested over a lengthy period of time may also accumulate toxic effects (Isabel et al. 2021)

Copper (Cu)

As shown in Table 1, the Cu concentration in the sample is the second highest after the Zn concentration of 0.56 ppm. Cu, like Zn, is also one of the essential elements, so this may be expected. This may be due to their participation in numerous biochemical and metabolic processes. Since Pantai Sepat is a well-established fishing spot, antifouling paint, fish landing stations, marine activities, and refuelling have been in use there for a long time. Moreover, because of its beauty, this location is becoming more well-known and attracting more tourists to the shore. There are numerous water-based activities, such as angling, swimming, boating, and banana boating. In addition, people are beginning to make a living by renting ATVs all along the shore, which contributes to both human-caused and heavy metal pollution.

As shown in Table 2, the measured concentration of Cu in the firm clams does not exceed the safety limit of 30.00 ppm. It is evident that the captured hard clams do not contain excessive levels of heavy metal pollutants. However, excessive Cu exposure in humans can cause acute toxicity and health problems due to impairment of liver and renal function (Chuan & Ibrahim 2017).

Compared to Liu et al. (2019), Cu's concentration is the second highest among the eight analysed elements. According to the study, the storage of excess Cu and Zn in oxygen- and nitrogen-bonded complexes by bivalves has enabled them to detoxify the metals by compartmentalizing them within membrane-limited vesicles of haemoglobin. In addition, Liu et al. (2019) also demonstrated that Cu or Zn accumulation in bivalves promotes the

absorption of other toxic metals, including Cd and Hg. These metal interactions may explain why bivalves have such high concentrations of Cd, Hg, Ni, and Pb.

Cadmium (Cd)

In this investigation, the Cd concentration in the *Meretrix* genus is lower than the Cu and Zn concentrations. According to Table 1, the concentration is only 0.06 parts per million. Since the research region is remote from industrial and agricultural sites, the marsh mollusc has a low concentration of cadmium. This is corroborated by the 2017 study by Chuan and Ibrahim (2017), in which the sampling region is located far from industrial and agricultural sites. In the investigation, the Cd concentration in the clams was lower than that of other elements observed.

The measured concentration of Cd (0.06 ppm) in the collected *Meretrix* genus is below the safety limit of 1.00 ppm, as shown in Table 2. Cadmium is a harmful element that enters the body systems of organisms, so even if the concentration is minimal, it should still be considered hazardous. Despite not being required for metabolic processes, cadmium is one of the environmental toxins that can cause considerable damage to human health, even at low concentrations. Long-term consumption of cadmium may result in toxicological risks and heavy metal-related diseases such as Parkinson's disease (Chuan & Ibrahim 2019; Dabwan & Taufiq 2016).

Lead (Pb)

According to Table 1, the concentration of Pb in the hard mussels is the lowest among the others, measuring 0.04 ppt. According to Chuan and Ibrahim (2017), the combustion of fossil fuels while on watercraft, as well as the use of diesel fuel and engine fuel, can contaminate the water with lead. Moreover, Pb is one of the most prevalent heavy metal environmental contaminants, especially in regions where metals tend to accumulate (Lichtfouse, Schwarzbauer & Robert 2013). In addition, the nearby automobile emissions from the combustion of fossil fuels may contribute to the release of lead into the aquatic environment, where it accumulates in sediments and organisms. Pantai Sepat is well-known for activities such as ATV riding along the beach, and people frequent the beach, particularly on weekends.

According to Dwi Harsono et al. (2017), the tourism burden is related to the increase in Pb in the clams. In the study by Dabwan and Taufiq (2016), the concentration of Pb in the oysters was the second highest out of the four trace elements measured. This is because the sampling area is close to the industrial site in which industrial effluents are deposited into the river and then into the ocean. In contrast, Kuantan's Pantai Sepat is a fishing village located far from any industrial or agricultural areas. Thus, the concentration is lower than in the previous study, and Pb may only be a result of boating while consuming fossil fuels. According to Table 2, the concentration of lead in the collected shellfish is 0.04 ppm, which is below the permissible limit. The safety threshold is 2.0 ppm. Therefore, the *Meretrix* genus collected from Pantai Sepat is contaminated minimally with lead. However, a minimal Pb intake over an extended period may also contribute to Pb accumulation in the body systems of consumers. Therefore, the consumption of mussels should be monitored periodically. Although these bivalves are seasonal and may not be consumed as frequently as other bivalves, it is much safer to exercise caution when handling them. A high concentration of lead may contribute to neurological disorders, such as interstitial nephritis and mental retardation, in adults and children. Long-term consumption also results in hypertension and cardiovascular disease for coastal consumers (Soisungwan et al. 2020).

Conclusion

The present study successfully assessed the amounts of certain heavy metals in *Meretrix* spp. The metals in the collected *Meretrix* spp. are arranged in the following order as such zinc (Zn), copper (Cu), cadmium (Cd), and lead (Pb). The amounts of these elements in the hard shells are found to be within the permissible limits set by the Malaysia Food Act (1983) and Food Regulation (1985) and the World Health Organisation (1982). Therefore, it can be concluded that the consumption of hard clams sourced from Pantai Sepat, Kuantan is considered to be safe.

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